

K&amp;S Case 4 - Prelim Amend

**BEST AVAILABLE COPY****Amendments to the Claims:**

*This listing of claims will replace all prior versions and listings of claims in the application.*

**Listing of Claims:**

Claim 1. (previously submitted) A method for detection and identification of constituents of extracts from plants or animals, natural or synthetic sources possessing medicinal value, using chromatographic fingerprinting techniques, the method comprising the steps of:

- i. extracting organic or organo-metallic compounds from plants or animal, natural or synthetic sources using a solvent;
- ii. subjecting the extract obtained in step i, to separating based on pH and polarity, using High Pressure Liquid Chromatography (HPLC)/ultra violet-visual (UV-VIS) 3-D chromatography technique;
- iii. generating contour and 3-D chromatograms of the constituents eluted in step ii, based on pH and polarity;
- iv. converting the 3-D and contour chromatogram obtained into a colored image, analyzing the colored image for its individual colors

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- using the co-ordinates denoting all its 3-dimension properties of said image;
- v. denoting the concentrations of the various constituents eluted with time;
- vi. generating a chromatogram based on color analyzed, having peaks at various retention times along with conjugative properties of the constituents;
- vii. identifying the compounds in said Ingredients by the Ultra Violet and Visible electromagnetic radiation absorptive properties of the various constituents in the image, thus creating a chromatographic fingerprint of the extract;
- viii. identifying, determining and classifying the compounds eluted as polar, medium polar and less or non-polar based on the polarity and conjugative properties;
- ix. generating a barcode for a selected peak using the X-axis as Retention Time, the Y-axis as Wavelength, R as the number of Red Pixels, G as the number of Green Pixels and B as the number of Blue Pixels; and
- x. generating a database of fingerprints and barcodes and identifying

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the respective compounds of the extract.

**Claim 2. (original)** The method as claimed in claim 1, wherein the solvents with different polarities are used for extraction based on the hydrophilic and hydrophobic nature of the constituents present in the sample under study, and ethyl alcohol is used as a solvent for preparation and for standardization of medicinal extracts.

**Claim 3. (original)** The method as claimed in claim 1, wherein the fingerprints are developed for the same medicinal extract under different pH ranges.

**Claim 4. (original)** The method as claimed in claim 1, wherein the HPLC/Ultra violet-visual 3-D chromatography technique is used by employing any commercially available HPLC apparatus with the Photo Diode Array detector, preferably with a gradient or ternary system of pumps.

**Claim 5. (original)** The method as claimed in claim 1, wherein the method is carried out using standard analytical parameters like extraction with

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ethyl alcohol, maintaining a run time of 0-60 minutes, eluting with a mobile phase of acetonitrile along with a phosphate buffer having a pH in the range of 5.5-7.5, and an Ultra Violet and Visible detector having the electromagnetic radiation range of 200-800nm for fingerprinting, chemical and therapeutic standardization.

**Claim 6. (previously submitted)** The method as claimed in claim 1, wherein the separation of step ii is based on polarity and is performed with a solvent of known pH.

**Claim 7. (previously submitted)** The method as claimed in claim 1, wherein converting the contour chromatograms into a colored image consists of identifying the conjugative and polarity properties of the constituents of the medicinal extract under study by imaging and extraction of colors of the image and presenting as data chromatograms.

**Claim 8. (original)** The method as claimed in claim 1, further comprising assessing a therapeutic efficacy of a single or formulated medicinal extract using the quality of the constituents present in a particular

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polarity and Ultra Violet-Visible absorptive zone out of different therapeutic zones of the chromatographic fingerprint.

Claim 9. (original) The method as claimed in claim 1, wherein in step ix, the barcode for the selected peak or peaks or image is generated by a software.

Claims 10 through 19 (cancelled)

Claim 20. (previously submitted) The method as claimed in claim 1, further including the step of using chemical analysis of the constituents present in the extract under study and their conjugative and polarity properties to determine therapeutic efficacy.

Claim 21. (previously submitted) The method as claimed in claim 1, wherein said analysis of the colored image in step iv provides a quick identification of the actual profile of the compounds present in the medicine under use along with the therapeutic efficacy of the constituents.

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Claim 22. (previously submitted) The method as claimed in claim 1, wherein said step of analyzing the colored image of step iv utilizes a photo diode array detector (PDA) of said high pressure liquid chromatography of step ii. To provide a chromatographic finger printing of herbal medicines and formulations.

Claim 23. (cancelled)

Claim 24. (previously submitted) The method as claimed in claim 1, wherein in step vii, said chromatographic fingerprint of the extract defines the blue print of the constituents present in an herbal medicine or formulation for an assay and for quick identification of the medicine under study.

Claim 25. (cancelled)

Claim 26. (original) The method as claimed in claim 1, wherein the natural or synthetic sources are adulterated, substituted, contradictable and commercial food and drug samples.

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Claims 27 through 33. (cancelled)

Claim 34. (previously submitted) The method as claimed in claim 24, wherein step ix further includes preparing a database of fingerprints together with correlations with the therapeutic efficacy which can be used to form generalizations of the therapeutic efficacy of a particular group of plants, classified as a group for a particular disease or therapeutic classification.

Claims 35 through 48. (cancelled)

Claim 49. (previously submitted) The method as claimed in claim 22, wherein said chromatographic finger printing delineates the data of the spectral properties of the constituents arranged in a specific order of polarity present in the material having medicinal value for chemical and therapeutic standardization.

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